Physiological Monitoring in Diving Mammals

Peter L. Tyack, Andreas Fahlman, and Michael Moore
Woods Hole Oceanographic Institution
266 Woods Hole Road, MS #50
Woods Hole MA 02543
phone: (508) 289-2818 fax: (508) 457-2041 email: ptyack@whoi.edu
phone: (508) 289-3463 fax: (508) 457-2041 email: afahlman@whoi.edu
phone: (508) 289-3228 fax: (508) 457-2041 email: mmoore@whoi.edu

Warren Zapol and Richard Anderson
Departments of Anesthesia, Critical Care, Cardiology and Dermatology
Harvard Medical School at Massachusetts General Hospital
Boston, Massachusetts 02114
email: wzapol@partners.org
email: rranderson@partners.org

Award Number: N00014-10-1-0791

LONG-TERM GOALS

The objective with this study is to develop and calibrate an invasive data logger to measure muscle O_2 saturation in large, freely diving whales. We intend to use this data logger to measure muscle O_2 saturation and determine how blood flow to muscle is altered during diving. These data will be important to determine if muscle blood flow is reduced during diving, and important to estimate how the dive response affects muscle N_2 levels and the risk of decompression sickness (DCS).

OBJECTIVES

Recent necropsy reports suggested a link between mass stranding of beaked whales and the use of naval mid-frequency sonar. The whales experienced symptoms that were similar to those caused by inert gas bubbles in human divers. These reports have increased the concern that intense sources of anthropogenic sound, such as that created by military sonar or during seismic exploration, may harm marine animals, particularly certain species of deep diving whales. Primary issues have centered on direct auditory damage, resonance of gas-containing spaces, and increased risk of DCS due to alteration in dive physiology or behavior, or acoustic enhancement of bubble formation and growth.

The stranding events have fueled an intense non-governmental organizational (NGO) scrutiny of the complex relationship between ocean noise, bubble injury and marine mammal strandings (http://www.awionline.org/oceans/Noise/IONC/index.htm). During a workshop held in Baltimore, MD USA in April 2004 [1] it was concluded that ‘gas-bubble disease, induced in supersaturated tissue by a behavioral response to acoustic exposure, is a plausible mechanism for the morbidity and mortality seen in cetaceans associated with sonar exposure.’

Report Documentation Page

Form Approved
OMB No. 0704-0188

1. REPORT DATE **2010**	2. REPORT TYPE	3. DATES COVERED **00-00-2010 to 00-00-2010**

4. TITLE AND SUBTITLE **Physiological Monitoring in Diving Mammals**	5a. CONTRACT NUMBER 5b. GRANT NUMBER 5c. PROGRAM ELEMENT NUMBER
6. AUTHOR(S)	5d. PROJECT NUMBER 5e. TASK NUMBER 5f. WORK UNIT NUMBER
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) **Woods Hole Oceanographic Institution,266 Woods Hole Road,Woods Hole,MA,02543**	8. PERFORMING ORGANIZATION REPORT NUMBER
9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)	10. SPONSOR/MONITOR'S ACRONYM(S) 11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION/AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT **unclassified**	b. ABSTRACT **unclassified**	c. THIS PAGE **unclassified**	**Same as Report (SAR)**	**5**	

Standard Form 298 (Rev. 8-98)
Prescribed by ANSI Std Z39-18

One approach to study this problem has been through theoretical calculations of the plausible tissue and blood N_2 levels, and recent work suggests that beaked whales commonly experience end-dive N_2 levels that would cause a significant proportion of DCS cases in terrestrial mammals [2]. Model sensitivity analysis further suggested that level of the dive response (cardiac output and the blood flow distribution) strongly influence the N_2 levels in blood and tissue, and thereby DCS risk [2-5]. It is assumed that all breath-hold diving marine mammals experience a dive response while submerged, with a reduction in cardiac output and a re-distribution of blood to the core. But the precise knowledge of how deep-diving whales distribute blood flow has never been measured. Improving such knowledge would significantly enhance our ability to predict end-dive blood and tissue N_2 levels, and determine if deep diving whales are at risk of DCS.

APPROACH

This project is separated into two aims: A) Development of a new generation of tag/data logger for marine mammals that will contain a sensor to be implanted into the muscle. The logger will collect physiological data from muscle tissue in freely diving marine mammals. The sensor will be tested and calibrated in terrestrial mammals at Massachussetts General Hospital, Boston.; B) The data logger will be tested in freely diving marine mammals in the field, and muscle O_2 saturation data will be collected.

A) A near infrared spectrophotometer connected to a data logging device will be developed and used to measure oxymyoglobin/hemoglobin saturation in freely deep-diving whales (e.g. beaked whales, sperm whales). The unit will be developed based upon the successful construction of an oximeter used in Weddel seals [6].

A needle delivery device will be fabricated that will allow the optical probe to be implanted into the muscle. The needle will be inserted briefly through the skin and blubber into the muscle to insert the cable and probe. The flexible cable will allow the muscle to move freely, resulting in minimal discomfort. Inserting a needle into a freely diving marine mammal is an invasive procedure, but current sedation experiments have proven this method to be an accepted method [7]. Initial experiments on terrestrial mammals and stranded or by-caught (post-mortem) marine mammals will assess the impact of the implantation to minimize the potential for inflammation and hematoma [7].

B) The data logger will be tested on a variety of diving marine mammals over 2 field seasons. We aim to perform translocation experiments in Northern elephant seals (*Mirounga angustirostris*) with collaborators at University of California Santa Cruz (UCSC). This allows us to perform controlled field experiments and to determine if the data logger is able to collect the physiological data and to assure minimal tissue trauma.

We will implant the oximeter into the muscle of freely, deep diving whales (beaked and/or sperm whales) during 2 field seasons.

WORK COMPLETED

Aim 1: Our collaborators at MGH have begun development of the oximeter sensor. In the first year, they have tested a range of LEDs and reflectance sensors to assure that we will be able to detect differences in oxy- and de-oxy hemoglobin. The light source and reflectance sensor is similar to the concept successfully used by Dr. Zapol [6], and consist of two laser diodes, one operating at 753-754 nm and the other at 812-814 nm. Both diodes are placed to have a beam angle of ~30 º and placed ~ 1-

2 cm apart. A light sensor will measure the reflectance at the emitted wavelengths, allowing us to compute the O_2 saturation from the ratio of the two wavelengths.

In the second year, a graduate student will be recruited at MGH to work full time on this project through a 6 month training program, from November until April. At the end of this period, it is expected that MGH will have completed the optical probe including some mechanical testing. We anticipate that to have integrated the probe into the data logger by the middle of the 2^{nd} year allowing us to continue with aim 2 at the end of year 2.

We are collaborating with a company in New Zealand (Paxarms) to develop the device that will implant the optical probe and attach the data logger to the whales. Paxarms have considerable experience at building such devices and have previously collaborated with Dr. Michael Moore to build a sedation needle that has been used to deliver injectable drugs to entangled whales [7]. Paxarms have begun to develop the device and begun testing how to implant the device into the whales.
One item of concern is that there is remarkably little structure linking the blubber to the sub-dermal sheath in cetaceans. This allows the blubber to move laterally over muscle. A cable crossing the blubber/muscle interface will therefore experience shear forces of varying degree. In an attempt to assess the stress to the cable, Paxarms built a testing platform, consisting of a blubber sample glued to a sheet of foam (Fig. 1). One possibility we are considering is to anchor the probe within the muscle. However, before considering this approach we will make tests on blubber/muscle samples from stranded (post-mortem) animals to determine the effect in real tissue. It may be argued that mechanical properties of muscle and foam might be quite different. The muscle is more viscous and elastic than the foam. The viscosity may make the tissue sticky and hold on to the cable rather than letting it slip in and out of the tissue.

Figure 1 shows the testing platform that was used to determine the mechanical forces acting on the implanted cable. The top left panel shows the testing platform, consisting of a blubber sample glued to a foam pad (the muscle). The cable was inserted through the blubber and foam (top right). Maximum lateral movement showed that the cable moved in and out of the sample a maximum 4 cm.

We expect to have the first prototype of the subdermal needle delivery system ready for testing on post-mortem stranded whales during the initial 6 months of the 2nd year. The needle delivery system will first be tested on post-mortem stranded or bycaught seals, dolphins and harbor porpoises. The samples will be requested from our ongoing collaboration with the NOAA Observer Program.
We are discussing various options of how to integrate the sensor into an archival data logger. Some possibilities include integration into a Dtag, or a custom-built data logger from UFI or Paxarms. The decision depends on hardware requirements of the finished probe and the device that will implant the probe into the muscle.

Aim 2: In the first year we have applied for and received all the necessary IACUC approvals and permits that will enable us to perform the field work on elephant seal in California and to deploy the tag on whales.

We consider the Northern elephant seal to be a suitable species to perform the first tests. This species performs long and deep dives, similar to sperm and beaked whales. It is also possible to perform controlled translocation experiments in this species where the instruments can be retrieved when the animal returns to the beach within a few days of release. For the field experiments on elephant seals, we are planning experiments in collaboration with researchers at UCSC in Santa Cruz. We hope to perform the first tests at the end of the 2nd year.

For tag deployment on whales, we have developed collaboration with Norwegian colleagues at the Norwegian Defence Research Establishment in Horten and the Polar Institute in Tromsø, Norway. We are currently discussing the time-frame and logistics for this tagging effort and how to combine it with on-going work to minimize the logistical burden and to reduce animal impact.

RESULTS

Aim 1) We have showed that we are able to detect differences in blood oxygenation using LEDs and reflectance sensors. We have evaluated and tested the lateral movement betweel whale blubber and muscle to assess the mechanical forces acting on the opritcal probe.

Aim 2) We are planning the field experiments in year 2 and 3 and all the animal care protocols and permits have been approved.

IMPACT/APPLICATIONS

This work is intended to enhance our understanding of how the dive response alters muscle blood flow and metabolism in large, freely diving whales. The results will provide information that will enable more realistic predictions of how the dive response varies during breath-hold diving at different activities. The study will also provide a new generation of data loggers that are able to collect physiological data in large whales with minimal impact.

Results from the completed study will help to improve our understanding about the physiology of marine mammals and improve modeling efforts that are aimed at estimating inert gas levels in breath-hold divers. The results can be used to determine how changes in dive behavior, from playback studies that measures avoidance patterns in deep diving whales, affect blood and tissue P_{N_2} levels. Thus, our results will enhance the fundamental understanding, interpretation and avoidance of the effect of

anthropogenic sound, and enable knowledgeable decisions about sonar deployment, related training exercises and responses to NGO concerns. This should be of value to the US Navy Marine Mammal Program.

REFERENCES

1. Cox, T.M., et al., *Understanding the impacts of anthropogenic sound on beaked whales.* Journal of Cetacean Research and Management, 2006. **7**: p. 177-187.
2. Hooker, S.K., R.W. Baird, and A. Fahlman, *Could beaked whales get the bends? Effect of diving behaviour and physiology on modelled gas exchange for three species: Ziphius cavirostris, Mesoplodon densirostris and Hyperoodon ampullatus.* Respiratory Physiology and Neurobiology, 2009. **167**: p. 235-246.
3. Fahlman, A., et al., *Estimating the effect of lung collapse and pulmonary shunt on gas exchange during breath-hold diving: the Scholander and Kooyman legacy* Respiratory Physiology and Neurobiology, 2009. **165**(28-39).
4. Fahlman, A., et al., *Deep diving mammals: Dive behavior and circulatory adjustments contribute to bends avoidance.* Respiratory Physiology & Neurobiology, 2006. **153**(1): p. 66-77.
5. Fahlman, A., et al., *To what extent does* N_2 *limit dive performance in king penguins?* Journal of Experimental Biology, 2007. **210**: p. 3344-3355.
6. Guyton, G.P., et al., *Myoglobin saturation in free-diving Weddell seals.* Journal of Applied Physiology, 1995. **79**: p. 1148-1155.
7. Moore, M., et al., *Sedation at Sea of Entangled North Atlantic Right Whales (Eubalaena glacialis) to Enhance Disentanglement.* PLoS ONE, 2010. **5**(3): p. e9597.